THE KEY NEUROENDOCRINE REGULATORS OF THE ONSET OF PUBERTY IN THE ATLANTIC BLUEFIN TUNA (*Thunnus thynnus*)

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Recently, significant progress on spawning induction in captive bluefin tuna (BFT, *Thunnus thynnus*), has been achieved providing the basis for the species' domestication. To further promote the development of a self- sustained BFT aquaculture, we investigated first sexual maturity in BFT reared from an immature stage in captivity. Accordingly, our major objectives were to evaluate: (i) maturational status of the brain-pituitary-gonadal (BPG) axis, and (ii) responsiveness of the BPG to exogenous hormones. Special emphasis was given to characterize the gonadotropins follicle stimulating hormone (FSH) and luteinizing hormone (LH) that act as central regulators of gonadal development and gamete maturation.

The growth parameters recorded for the captive BFT juveniles are consistent with the length-weight relationship established for wild Mediterranean BFT stocks. The histological analyses of the gonads indicate advanced sexual maturation in BFT males

compared to females, yet it is not yet clear whether this phenomenon typifies wild stocks or is induced due to the culture conditions. The hormone measurements show expression and accumulation of both gonadotropins in the pituitaries of immature and mature BFT. The pituitary LH content increased concomitantly with the age of the fish, exhibiting sex dimorphic patterns (i.e. 3-fold higher levels in females) in adult but not in juvenile BFT. The pituitary FSH levels, however, were elevated in 2Y immature males and in fully mature adults. Comparable to mammals, the intra-pituitary FSH/LH ratio was found to be higher (>1) in sexually immature than in maturing or pubertal BFT. Nevertheless, in the 3Y BFT females, which were all immature, the onset of puberty appears to require some other prerequisites, such as a rise in the LH storage above a minimal threshold. Our *in vitro* trials further demonstrated the capacity of rFSH and to a lesser extent that of rLH to stimulate cell proliferation in the immature ovarian and testicular fragments. Both rFSH and rLH have failed to stimulate steroidogenesis, yet pre-treatment with KiSS containing EVAc implants appeared to potentiate FSH-stimulated steroidogenesis in the immature testes. On the other hand, the expression levels of both the GtH-R and IGF I genes in the testicular fragments, derived from BFT juveniles and further exposed to the rLH treatment, showed dose-dependent pattern.

Future studies testing the effects of captivity and hormone-based treatments on precocious maturity at relatively small body size are expected to facilitate the handling in confined environments, and to greatly improve the cost-efficiency of BFT farming.